

**WHAT IS CLAIMED IS:**

1           1.       A method of mapping the location of a post-translational modification  
2 of a post-translationally modified peptide, said method comprising:

3           (a) contacting said peptide with a chemical modification reagent that converts  
4 a post-translationally modified amino acid residue of said peptide into a substrate for a  
5 peptidase, thereby producing a chemically modified peptide comprising a chemically  
6 modified amino acid residue;

7           (b) contacting said chemically modified peptide with said peptidase under  
8 conditions appropriate to degrade said chemically modified peptide, thereby producing a  
9 degraded chemically modified peptide; and

10          (c) querying said degraded chemically modified peptide to ascertain said  
11 location of said post-translational modification.

1           2.       The method of claim 1, further comprising:

2           (d) prior to step (a), contacting a substrate amino acid of said peptide that is a  
3 natural substrate for said peptidase with a blocking agent thereby converting said substrate  
4 amino acid into a side-chain protected amino acid that is not a substrate for said peptidase.

1           3.       The method of claim 2, wherein said substrate amino acid is a lysine,  
2 wherein said blocking agent converts said lysine into a side-chain protected lysine selected  
3 from the group consisting of a carbamate, an amide, an N-sulfonyl, an N-sulfenyl, an N-nitro,  
4 an N-nitroso, an N-oxide, an imine, an N-alkyl amine, an N-aryl amine, an N-phosphinyl, an  
5 N-phosphoryl, and an enamine .

1           4.       The method of claim 2, wherein said side chain protected lysine is  
2 selected from the group consisting of Lys(Aloc), Lys(Ac), Lys(Boc), Lys(biotinyl), Lys(2-  
3 bromo-Z), Lys(2-chloro-Z), Lys(Dnp), Lys(Fmoc), Lys(For), Lys(Me)<sub>2</sub>, Lys(nicatinoyl),  
4 Lys(Tfa), Lys(Tos), Lys(Z), Lys(Z)(isopropyl), Lys(Boc)(isopropyl), Lys(dansyl), Lys(Dde),  
5 Lys(Me)<sub>3</sub>, Lys(Mtt), Lys(palitoyl), Lys(TNM), Lys(acetimidoyl), Lys(2,4,-dichloro-Z),  
6 Lys(Me), Lys(p-nitro-Z), Lys(5/6 FAM), Lys(pyrenebutyryl), and Lys(guanidinyl).

1           5.       The method of claim 1, wherein said substrate amino acid is aspartic  
2 acid, wherein said blocking agent converts said aspartic acid into a side-chain protected  
3 aspartic acid selected from an ester, an amide, an oxalose, an oxazolines, a stannyl ester, and  
4 an hydrazide.

1                   6.     The method of claim 1, wherein side chain protected aspartic acid is  
2 selected from the group consisting of Asp(OBzl), Asp(OcHex), Asp(OtBu), Asp(OMpe),  
3 Asp(Ofm), Asp(Osu), Asp(2-phenylisopropyl ester), and Asp(ONp).

1                   7.     The method of claim 1, wherein said peptidase is selected from the  
2 group consisting of a serine endopeptidase, a metalloendopeptidase, a cysteine  
3 endopeptidase, and an aspartic endopeptidase.

1                   8.     The method of claim 1, wherein said peptidase is a lysine-specific  
2 peptidase.

1                   9.     The method of claim 8, wherein said lysine-specific peptidase is  
2 selected from the group consisting of endoproteinase Lys-C, lysyl endopeptidase, trypsin,  
3 plasma kallikrein, oligopeptidase B, tryptase, plasmin, acrosin, granzyme A, yapsin 1,  
4 peptidyl-Lys metalloendopeptidase, and magnolsyin.

1                   10.    The method of claim 8, wherein said lysine-specific peptidase is  
2 selected from the group consisting of endoproteinase Lys-C, lysyl endopeptidase and trypsin.

1                   11.    The method of claim 1, wherein said peptidase is an aspartate-specific  
2 peptidase.

1                   12.    The method of claim 11, wherein said aspartate-specific peptidase is  
2 selected from peptidyl-aspartate metalloendopeptidase and nepenthesin.

1                   13.    The method of claim 1, wherein said querying comprises mass  
2 spectrographic detection of said chemically modified amino acid residue of said degraded  
3 chemically modified peptide.

1                   14.    The method according to claim 1, further comprising:  
2 (e) prior to step (a), contacting said peptide with an elimination reagent that  
3 causes the elimination of a post-translationally added substituent of said post-translationally  
4 modified amino acid residue.

1                   15.    The method of claim 14, wherein said post-translationally modified  
2 amino acid residue is selected from the group consisting of a post-translationally modified  
3 serine and a post-translationally modified threonine.

1                   16.     The method of claim 14, wherein said post-translationally modified  
2 amino acid residue is a phosphorylated amino acid residue.

1                   17.     The method according to claim 14, wherein said elimination is a  $\beta$ -  
2 elimination giving rise to an alkene moiety.

1                   18.     The method according to claim 1, wherein said modification reagent  
2 reacts with said post-translationally modified amino acid residue via a Michael addition.

1                   19.     The method of claim 18, wherein said modification reagent is selected  
2 from the group consisting of sodium sulfate and cysteamine.

1                   20.     A reactive solid phase material comprising:  
2                   (a) a solid support; and  
3                   (b) a solid support reactive moiety immobilized on said solid support, wherein  
4 said solid support reactive moiety is reactive towards a synthetically modified amino acid  
5 residue of a post-translationally modified peptide, said synthetically modified amino acid  
6 residue produced by elimination a post-translationally added substituent of said post-  
7 translationally modified peptide.

1                   21.     The material according to claim 20, wherein said synthetically  
2 modified amino acid residue comprises an alkene moiety.

1                   22.     A method of immobilizing a post-translationally modified peptide  
2 comprising a post-translationally modified amino acid, said method comprising:  
3                   (i) contacting said peptide with an elimination reagent that causes the  
4 elimination of a post-translationally added substituent of said post-translationally modified  
5 amino acid residue thereby producing a synthetically modified amino acid;  
6                   (ii) reacting said synthetically modified amino acid with a reactive solid phase  
7 material thereby immobilizing said post-translationally modified peptide, said reactive solid  
8 phase material comprising:  
9                   (a) a solid support; and  
10                   (b) a solid support reactive moiety immobilized on said solid support,  
11                   wherein said solid support reactive moiety is reactive towards said  
12 synthetically modified amino acid residue.